

Amendments to the Claims

This listing of claims will replace all prior versions, and listings, of claims in the application:

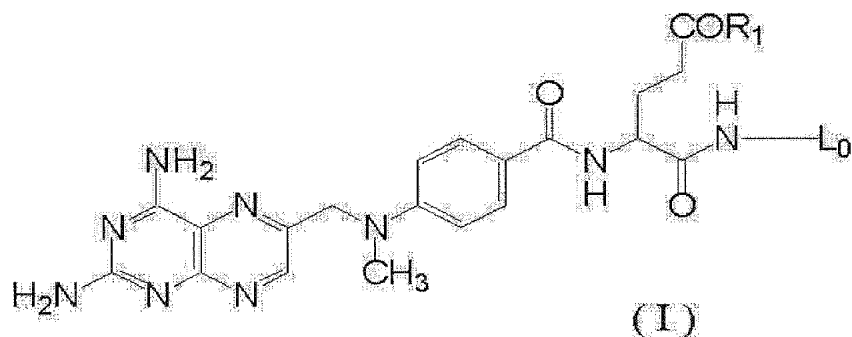
Listing of Claims:

1. (Currently Amended) A hyaluronic acid-methotrexate conjugate, wherein methotrexate is conjugated with a carboxyl group of hyaluronic acid, a hyaluronic acid derivative, or a salt thereof through a linker containing a peptide chain consisting of 1 to 8 amino acids; or a salt of the conjugate.
2. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1, wherein the linker contains a peptide chain consisting of 1 to 8 amino acids and a C₂₋₂₀ alkylenediamine chain, wherein the alkylenediamine chain optionally has 1 to 5 oxygen atoms inserted thereinto and/or is optionally substituted by a carboxyl group or a C₁₋₆ alkoxycarbonyl group.
3. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1 or 2, wherein the conjugation rate of methotrexate is 0.5% to 4.5% based on the total carboxyl groups of hyaluronic acid.

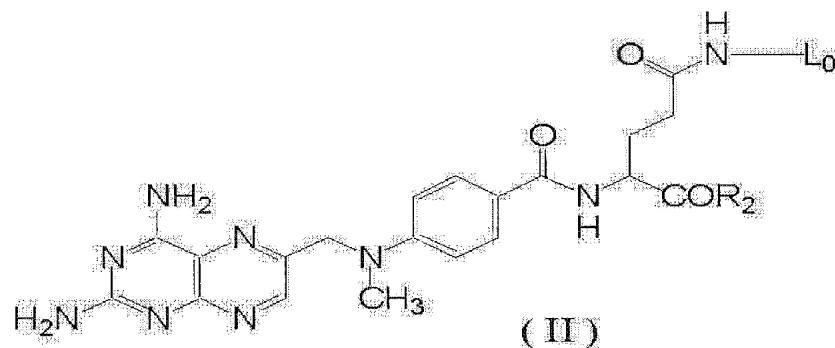
4. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1, wherein the molecular weight of hyaluronic acid is 600,000 daltons or more.

5. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1, wherein methotrexate conjugated with the linker is represented by formula (I), (II), (III), or (IV):

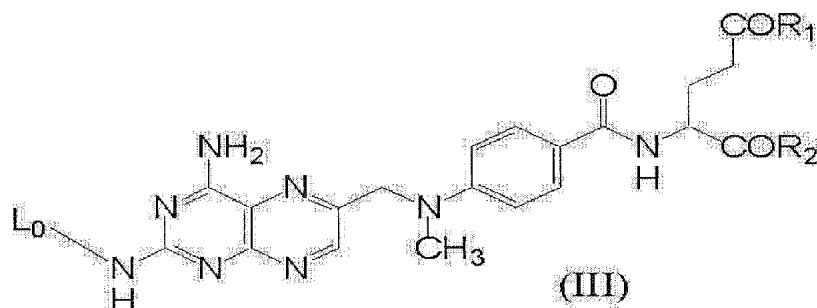
[Formula 1]



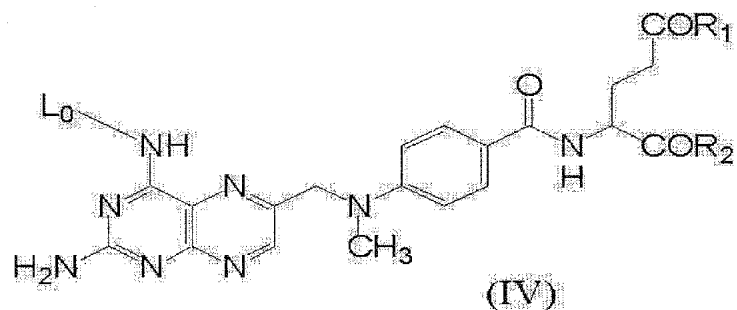
[Formula 2]



[Formula 3]



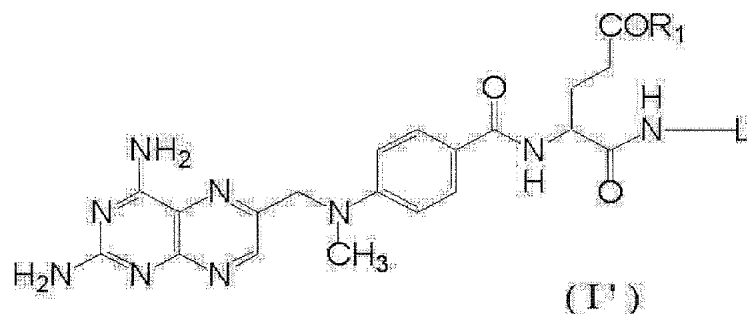
[Formula 4]



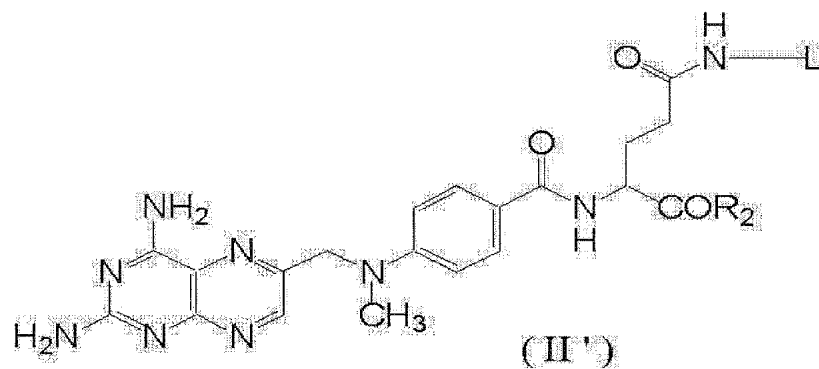
wherein R₁ and R₂ are each independently a hydroxy group, an amino group, a C₁₋₆ alkoxy group, a C₁₋₆ alkylamino group, or a di-C₁₋₆ alkylamino group; L₀ is the conjugation position of the linker.

6. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1, wherein the linker containing a peptide chain and methotrexate conjugated with the linker is represented by formula (I') or (II'):

[Formula 5]



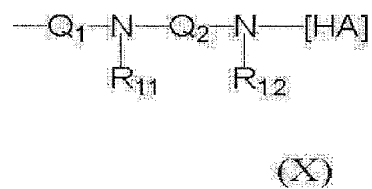
[Formula 6]



wherein R₁ and R₂ are each independently a hydroxy group, an amino group, a C₁₋₆ alkoxy group, a C₁₋₆ alkylamino group, or a di-C₁₋₆ alkylamino group;

L is a linker represented by formula (X):

[Formula 7]



wherein Q₁ forms, together with -NH- binding thereto, a peptide chain consisting of 1 to 8 amino acids; residues of amino acids contained in the peptide chain are each independently optionally substituted or protected by one or more groups selected from the group consisting of a C₁₋₆ alkyl group, a C₁₋₆ alkylcarbonyl group, a C₁₋₆ alkoxy carbonyl group, a formyl group, a C₁₋₆ alkylsulfonyl group, and a C₆₋₁₀ arylsulfonyl group; amide bonds contained in the peptide chain are each independently optionally substituted on the nitrogen atom by one or more C₁₋₆ alkyl groups and/or C₁₋₆ alkylcarbonyl groups; and carboxyl groups contained in the residues are each independently optionally converted to an amide group optionally substituted by one or two C₁₋₆ alkyl groups;

R₁₁ and R₁₂ are each independently a hydrogen atom or a C₁₋₆ alkyl group;

Q₂ is C₂₋₂₀ alkylene, wherein the alkylene optionally has 1 to 5 oxygen atoms inserted thereinto and/or is optionally substituted by a carboxyl group or a C₁₋₆ alkoxy carbonyl group; and

[HA] represents the position of conjugation with the hyaluronic acid, derivative, or salt thereof, and the linker forms an amide bond with a carboxyl group contained in the hyaluronic acid, derivative or salt thereof.

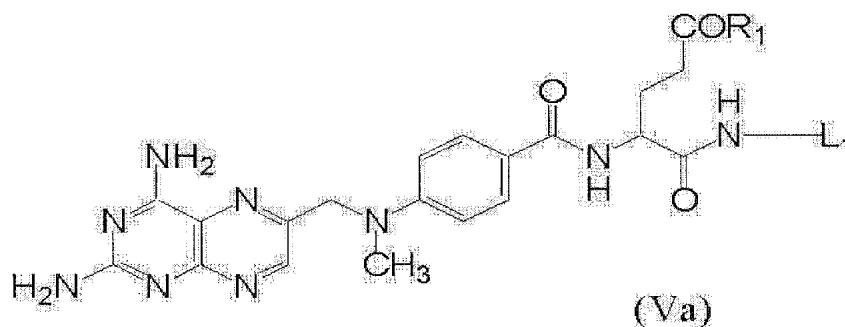
7. (Currently Amended) A pharmaceutical composition containing the hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1 as an active ingredient.

8. (Currently Amended) A therapeutic drug for joint diseases, containing the hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1 as an active ingredient.

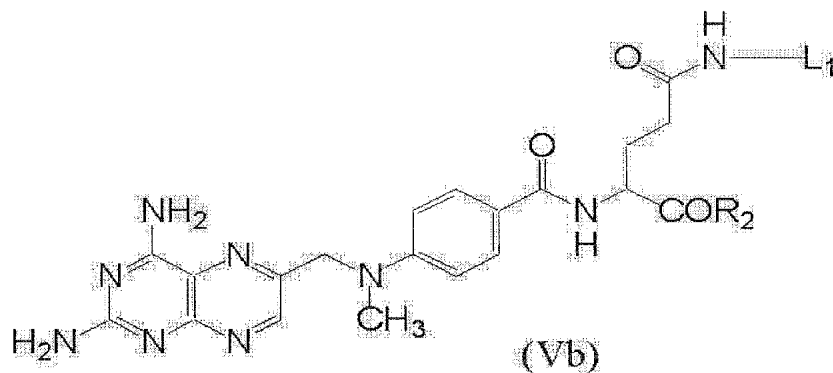
9. (Original) The therapeutic drug for joint diseases according to claim 8, which is a topical preparation for administration into the joint.

10. (Currently Amended) A compound of formula (Va) or (Vb):

[Formula 8]



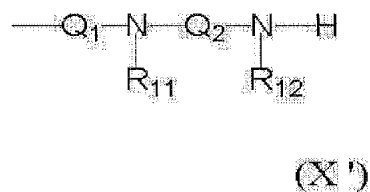
[Formula 9]



wherein R_1 and R_2 are each independently a hydroxy group, an amino group, a C_{1-6} alkoxy group, a C_{1-6} alkylamino group, or a di- C_{1-6} alkylamino group;

L_1 is a linker represented by formula (X'):

[Formula 10]



wherein Q_1 forms, together with —NH— binding thereto, a peptide chain consisting of 1 to 8 amino acids; residues of amino acids contained in the peptide chain are each independently optionally substituted or protected by one or more groups selected from the group consisting of a C_{1-6} alkyl group, a C_{1-6} alkylcarbonyl group, a C_{1-6} alkoxy carbonyl group, a formyl group, a C_{1-6} alkylsulfonyl group, and a C_{6-10} arylsulfonyl group; amide bonds contained in the

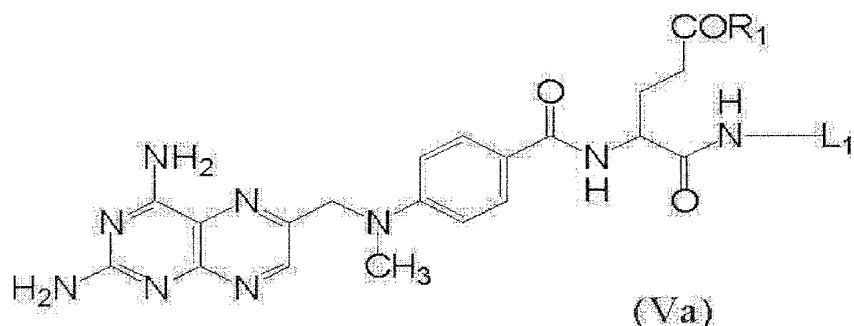
peptide chain are each independently optionally substituted on the nitrogen atom by one or more C₁₋₆ alkyl groups and/or C₁₋₆ alkylcarbonyl groups; and carboxyl groups contained in the residues are each independently optionally converted to an amide group optionally substituted by one or two C₁₋₆ alkyl groups;

R₁₁ and R₁₂ are each independently a hydrogen atom or a C₁₋₆ alkyl group; and

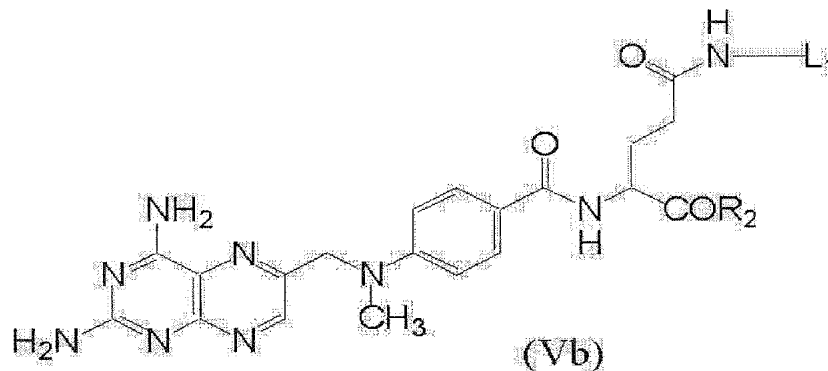
Q₂ is a C₂₋₂₀ alkylene, wherein the alkylene optionally has 1 to 5 oxygen atoms inserted thereinto and/or is optionally substituted by a carboxyl group or a C₁₋₆ alkoxycarbonyl group.

11. (Currently Amended) A process for producing the hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 1, which comprises the steps of reacting the compound of formula (Va) or (Vb) with hyaluronic acid, a hyaluronic acid derivative, or a salt thereof, and converting a carboxyl group of the hyaluronic acid, derivative, or salt thereof to an N-substituted amide group, wherein (Va) and (Vb) are as follows:

[Formula 8]



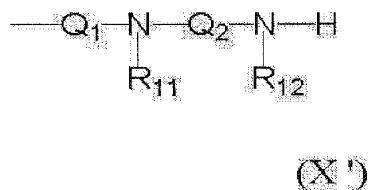
[Formula 9]



wherein R₁ and R₂ are each independently a hydroxy group, an amino group, a C₁₋₆ alkoxy group, a C₁₋₆ alkylamino group, or a di-C₁₋₆ alkylamino group;

L₁ is a linker represented by formula (X'):

[Formula 10]



wherein Q₁ forms, together with -NH- binding thereto, a peptide chain consisting of 1 to 8 amino acids; residues of amino acids contained in the peptide chain are each independently optionally substituted or protected by one or more groups selected from the group consisting of a C₁₋₆ alkyl group, a C₁₋₆ alkylcarbonyl group, a C₁₋₆ alkoxy carbonyl group, a formyl group, a C₁₋₆ alkylsulfonyl group, and a C₆₋₁₀ arylsulfonyl group; amide bonds contained in the

peptide chain are each independently optionally substituted on the nitrogen atom by one or more C₁₋₆ alkyl groups and/or C₁₋₆ alkylcarbonyl groups; and carboxyl groups contained in the residues are each independently optionally converted to an amide group optionally substituted by one or two C₁₋₆ alkyl groups;

R₁₁ and R₁₂ are each independently a hydrogen atom or a C₁₋₆ alkyl group; and

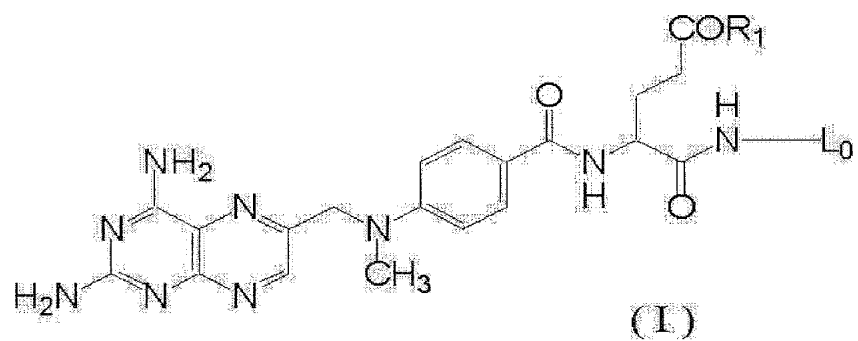
Q₂ is a C₂₋₂₀ alkylene, wherein the alkylene optionally has 1 to 5 oxygen atoms inserted thereinto and/or is optionally substituted by a carboxyl group or a C₁₋₆ alkoxycarbonyl group.

12. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 2, wherein the molecular weight of hyaluronic acid is 600,000 daltons or more.

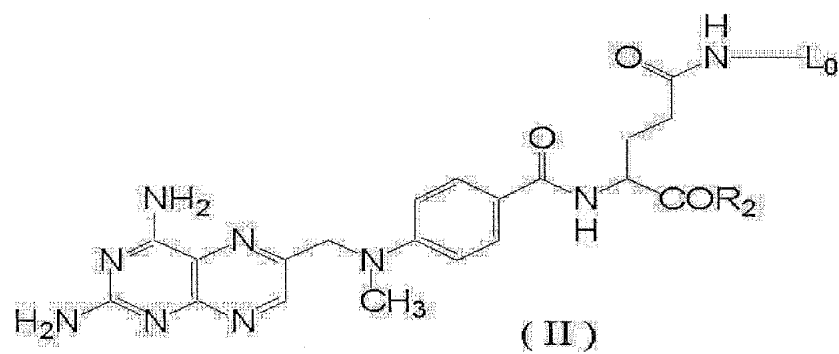
13. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 3, wherein the molecular weight of hyaluronic acid is 600,000 daltons or more.

14. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 2, wherein methotrexate conjugated with the linker is represented by formula (I), (II), (III), or (IV):

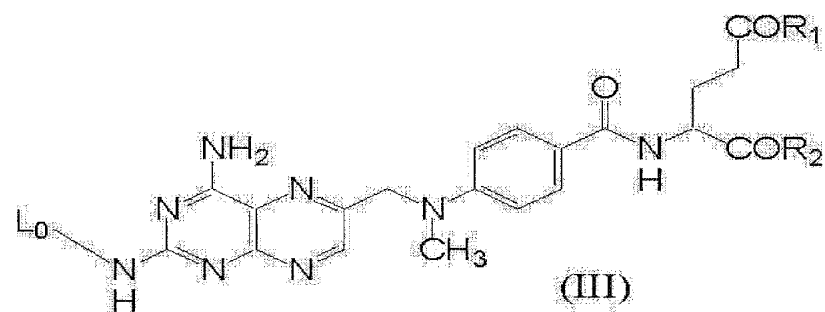
[Formula 1]



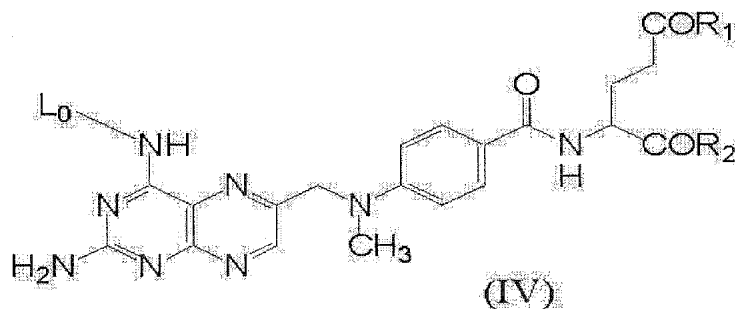
[Formula 2]



[Formula 3]



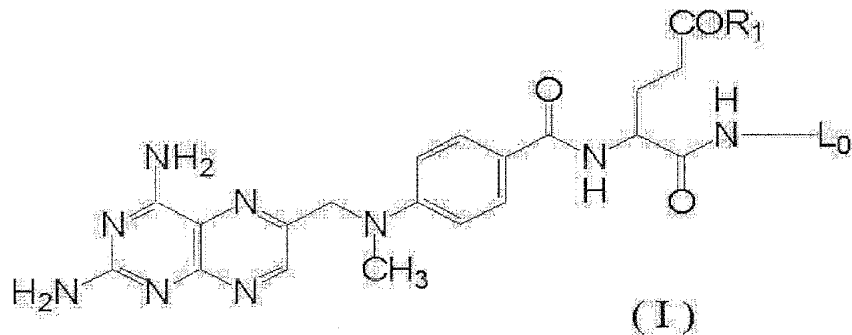
[Formula 4]



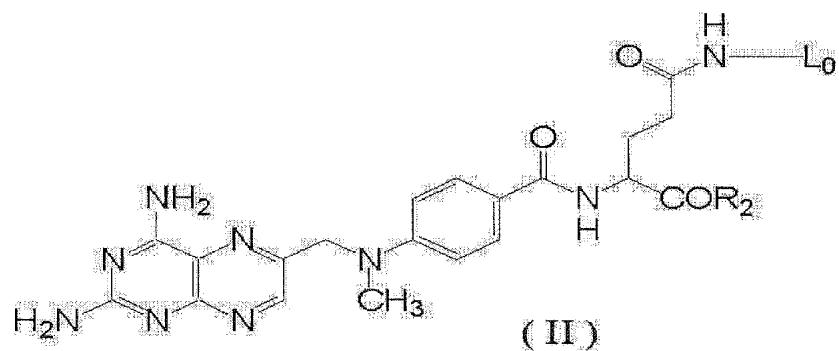
wherein R_1 and R_2 are each independently a hydroxy group, an amino group, a C_{1-6} alkoxy group, a C_{1-6} alkylamino group, or a di- C_{1-6} alkylamino group; L_0 is the conjugation position of the linker.

15. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 3, wherein methotrexate conjugated with the linker is represented by formula (I), (II), (III), or (IV):

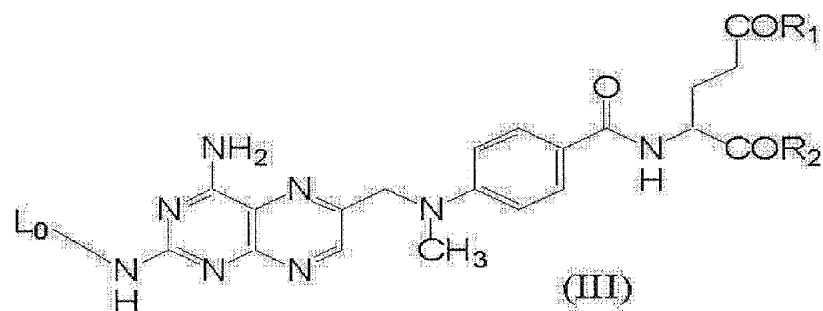
[Formula 1]



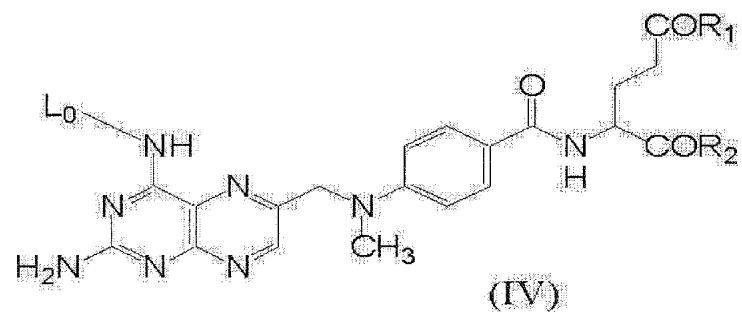
[Formula 2]



[Formula 3]



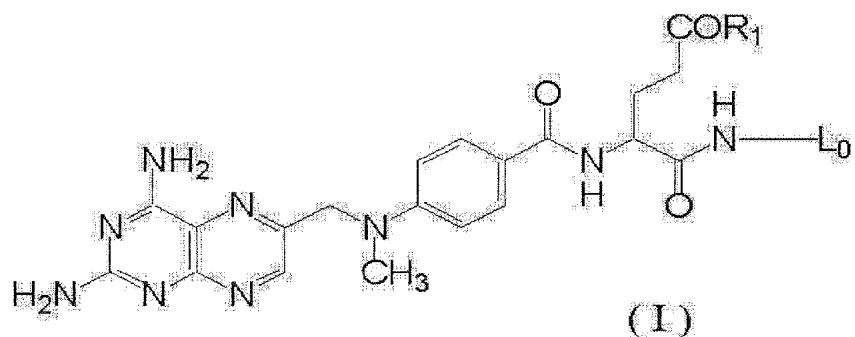
[Formula 4]



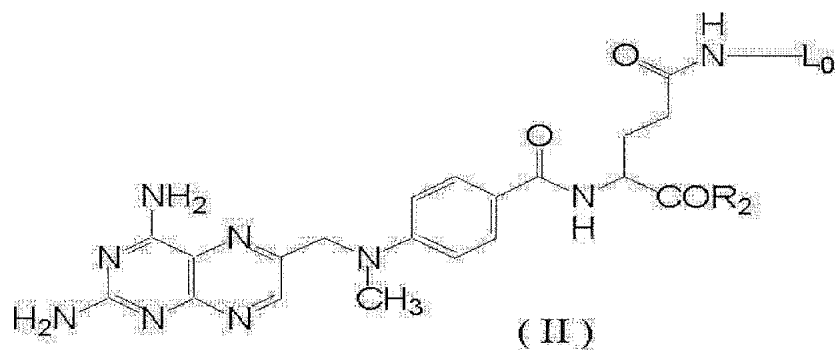
wherein R_1 and R_2 are each independently a hydroxy group, an amino group, a C_{1-6} alkoxy group, a C_{1-6} alkylamino group, or a di- C_{1-6} alkylamino group; L_0 is the conjugation position of the linker.

16. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 4, wherein methotrexate conjugated with the linker is represented by formula (I), (II), (III), or (IV):

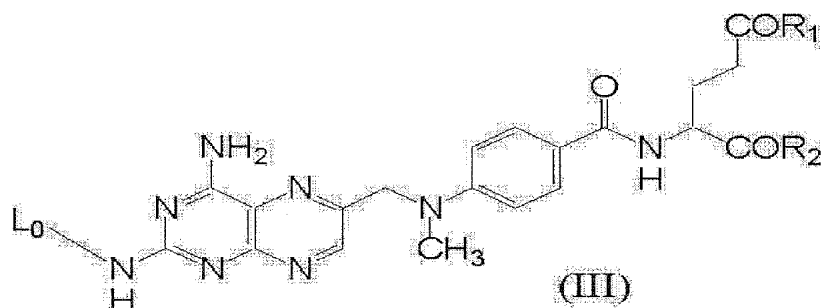
[Formula 1]



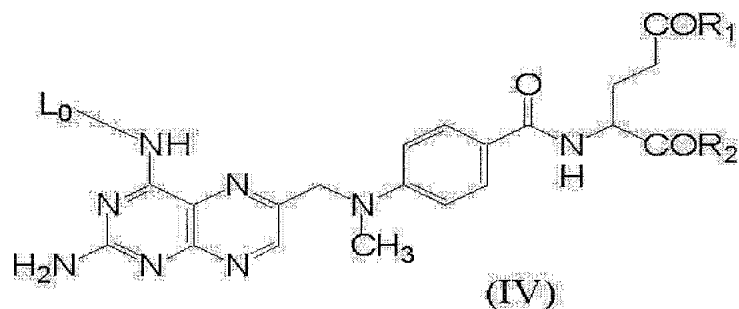
[Formula 2]



[Formula 3]



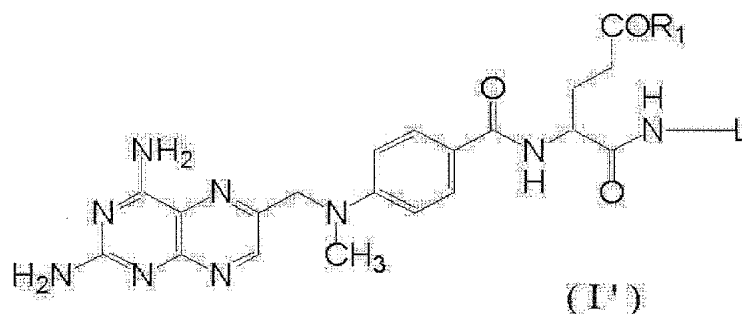
[Formula 4]



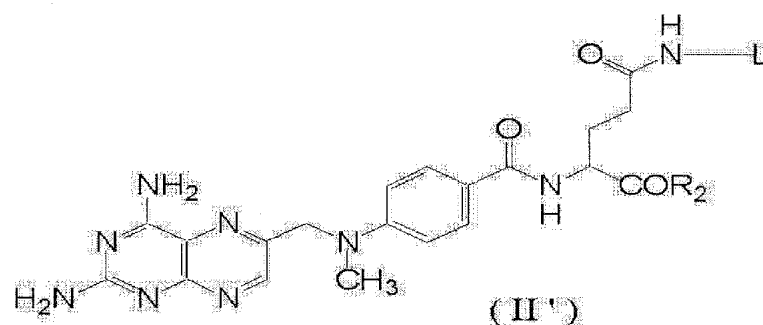
wherein R₁ and R₂ are each independently a hydroxy group, an amino group, a C₁₋₆ alkoxy group, a C₁₋₆ alkylamino group, or a di-C₁₋₆ alkylamino group; L₀ is the conjugation position of the linker.

17. (Currently Amended) The hyaluronic acid-methotrexate conjugate or the salt thereof according to claim 2, wherein the linker containing a peptide chain and methotrexate conjugated with the linker is represented by formula (I') or (II'):

[Formula 5]



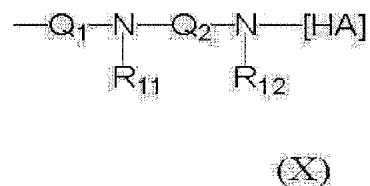
[Formula 6]



wherein R_1 and R_2 are each independently a hydroxy group, an amino group, a C_{1-6} alkoxy group, a C_{1-6} alkylamino group, or a di- C_{1-6} alkylamino group;

L is a linker represented by formula (X):

[Formula 7]



wherein Q₁ forms, together with -NH- binding thereto, a peptide chain consisting of 1 to 8 amino acids; residues of amino acids contained in the peptide chain are each independently optionally substituted or protected by one or more groups selected from the group consisting of a C₁₋₆ alkyl group, a C₁₋₆ alkylcarbonyl group, a C₁₋₆ alkoxy carbonyl group, a formyl group, a C₁₋₆ alkylsulfonyl group, and a C₆₋₁₀ arylsulfonyl group; amide bonds contained in the peptide chain are each independently optionally substituted on the nitrogen atom by one or more C₁₋₆ alkyl groups and/or C₁₋₆ alkylcarbonyl groups; and carboxyl groups contained in the residues are each independently optionally converted to an amide group optionally substituted by one or two C₁₋₆ alkyl groups;

R₁₁ and R₁₂ are each independently a hydrogen atom or a C₁₋₆ alkyl group;

Q₂ is C₂₋₂₀ alkylene, wherein the alkylene optionally has 1 to 5 oxygen atoms inserted therein and/or is optionally substituted by a carboxyl group or a C₁₋₆ alkoxy carbonyl group; and

[HA] represents the position of conjugation with the hyaluronic acid, derivative, or salt thereof, and the linker forms an amide bond with a carboxyl group contained in the hyaluronic acid, derivative, or salt thereof.